Supporting Information

Crystal structures of R47H and WT TREM2 ECD

The two R47H TREM2 ECD molecules in the asu are nearly identical with an overall r.m.s.d. of 0.38 Å for backbone Cα atoms (residues 19- 130). The TREM2 R47H variant maintains an overall similar fold to other Ig-like family members with two sheets of anti-parallel β-strands. One β sheet is composed of strands A-G-F-C-C' and the other sheet of B-E-D-C" (Fig. 1a). Residues R76 to S81 in β-strand C" and loop C"-D are not modeled due to lack of electron density (shown in dotted black line Fig. 1a) and β-strand C" is absent in this structure. Most residues in the WT structure could be modeled into the electron density (except N-terminal residues 19-21 and C-terminal residues 131-141). The overall fold of the WT TREM2 ECD structure is almost identical to the recently reported structure at 3.1 Å (Fig. 1c) (15), with an overall r.m.s.d. of 0.50 Å. We were also able to model a single N-acetylglucosamine (NAG) glycan on residue N79 (Figs. 1b and 1c) in the WT structure. All six molecules in the asu are highly similar, with an average r.m.s.d. of the backbone Cα atoms among the 6 molecules of 0.26 Å (range: 0.21 Å – 0.32 Å). Chain D shows a poorer fit to the electron density and poorer geometry in general, indicating that this molecule is likely less ordered than the other chains in the asymmetric unit.

Table S1 Data collection and refinement statistics

	TREM2 WT	TREM2 R47H variant	TREM2 WT + phosphatidylserine
Data Collection			
Space group	$P\,4_1\,2_1\,2$	$P\,2_1\,2_1\,2$	$P\,4_1\,2_1\,2$
Cell dimensions			
a,b,c (Å)	160.6, 160.6, 86.6	23.8, 62.5, 125.3	160.3, 160.3, 86.2
α,β,γ(°)	90, 90, 90	90, 90, 90	90, 90, 90
Resolution range	45.54 - 2.2 (2.279 - 2.2)	34.73 - 1.8 (1.864 - 1.8)	47.36 – 2.2 (2.279 – 2.2)
Total reflections	115756 (11400)	35470 (3087)	114765 (11288)
Multiplicity	2.0 (2.0)	2.0 (1.9)	2.0 (2.0)
Completeness (%)	100 (100)	98 (91)	99 (100)
Mean I/sigma(I)	14.67 (1.57)	8.66 (2.50)	20.53 (1.90)
R-merge	0.0378 (0.490)	0.0499 (0.188)	0.0313 (0.405)
Refinement			
R-work/Rfree	0.218/0.250	0.206/0.246	0.218 /0.255
Number of non-hydrogen atoms	5475	1744	5429
macromolecules	5058	1631	5058
ligands	245	N. A.	288
solvent	172	113	83
RMS(bonds)(Å)	0.024	0.006	0.031
RMS(angles)((°))	1.06	1.13	2.25
Average B-factor	51.54	11.43	54.00
macromolecules	51.23	11.23	52.89
ligands	61.50	N.A.	76.50
solvent	46.44	14.31	44.19

Values in parentheses are for the highest resolution shell.

Table S1. Data collection and refinement statistics for human WT TREM2, R47H TREM2 variant, and PS-soaked WT TREM2.

Table S2 Apo WT TREM2 and PS-Bound WT TREM2 Interface Buried Accessible Surface Area (Å^2)

Molecule Interface	TREM2 WT Apo	TREM2 WT PS
A-B	672	690
A-C	675	715
B-C	687	708
D-A	476	462
D-B	559	644
E-B	472	438
E-C	672	573
F-C	472	451
F-A	618	621

Table S2. Interface buried accessible surface areas (Å^2) for apo WT TREM2 and PS-bound WT TREM2.

Table S3 Apo WT TREM2 and PS-Bound WT TREM2 Interface Shape Complementarity (Sc)

Molecule Interface	TREM2 WT Apo	TREM2 WT PS
A-B	0.677	0.673
A-C	0.697	0.660
B-C	0.728	0.744
D-A	0.622	0.580
D-B	0.810	0.700
E-B	0.632	0.533
E-C	0.683	0.791
F-C	0.595	0.589
F-A	0.745	0.683

Table S3. Shape complementarity calculations ($Å^2$) for apo WT TREM2 and PS-bound WT TREM2 molecular interfaces.

Table S4 TREM2 Constructs for Crystallography

Construct	Expression System
huTREM2 (1-174)::GSS::Flag::6xHis	293-S
huTrem2 (1-174) R47H::GSS::Flag::6xHis	293-S
huTrem2 (1-131)	E. coli refold
huTrem2 (1-131) R47H[1]	E. coli refold
huTREM2 (1-131)::TEV::6xHis	293-6E
huTrem2 (1-174)::TEV::6xHis	293-S
huTrem2 (1-174) R47H::TEV::6xHis	293-S
huTREM2 (1-174) N20D::TEV::6xHis[2]	293-S

[1]Construct for TREM2 R47H variant crystal structure
[2]Construct for WT TREM2 and WT TREM2/PS crystal structures

Table S4. TREM2 constructs for crystallography.

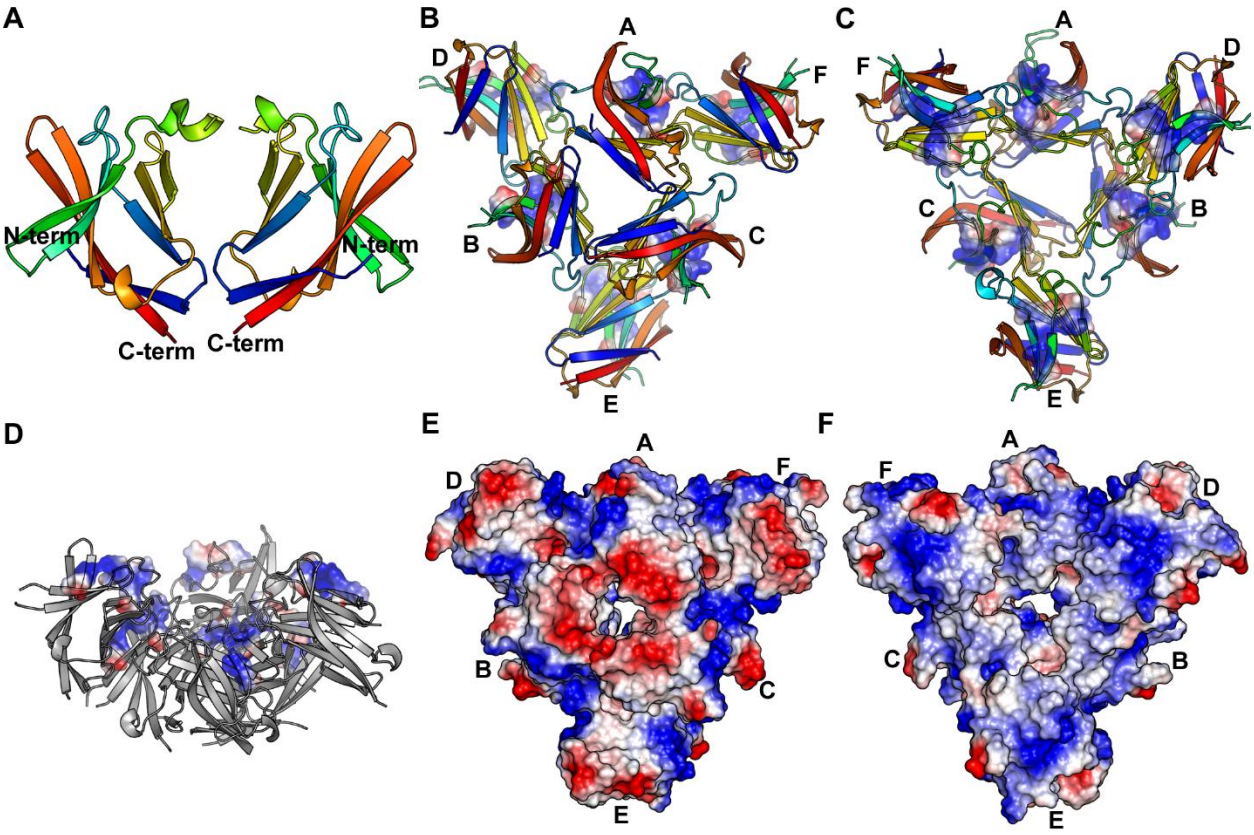

Figure S1. (a) Dimeric packing of 2 R47H TREM2 variant molecules in the asymmetric unit in 'tail-to-tail' interaction (rainbow). (b) Trimeric packing of 6 WT TREM2 molecules in the asymmetric unit with inner trimer oriented in a forward-facing manner and exterior neighboring molecules toward the posterior (rainbow cartoon). Electrostatic surface diagram of PLIS is indicated. CDR2 loop region is in green. (c) Trimeric packing of 6 WT TREM2 molecules in the asymmetric unit with exterior neighboring molecules oriented in a forward-facing manner and inner trimer toward the posterior (rainbow cartoon). Electrostatic surface diagram of PLIS is indicated. CDR2 loop region is in green. (d) Side orientation of WT TREM2 trimeric arrangement with C-termini of all molecules oriented downwards (grey cartoon). Electrostatic surface of PLIS is indicated. (e) Electrostatic surface diagram of trimeric packing of 6 WT TREM2 molecules in the asymmetric unit with inner trimer oriented in a forward-facing manner and exterior neighboring molecules toward the posterior. (f) Electrostatic surface diagram of trimeric packing of 6 WT TREM2 molecules in the asymmetric unit with exterior neighboring molecules oriented in a forward-facing manner and inner trimer toward the posterior.

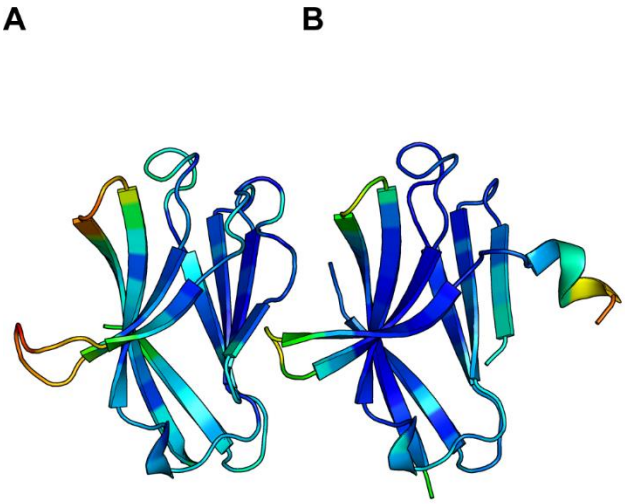

Figure S2. (a) WT TREM2 and (b) R47H TREM2 structures depicted in cartoon with associated B-factors in gradient coloring. Low B-factor values trend towards cooler coloring; high B-factor values trend towards warmer coloring.

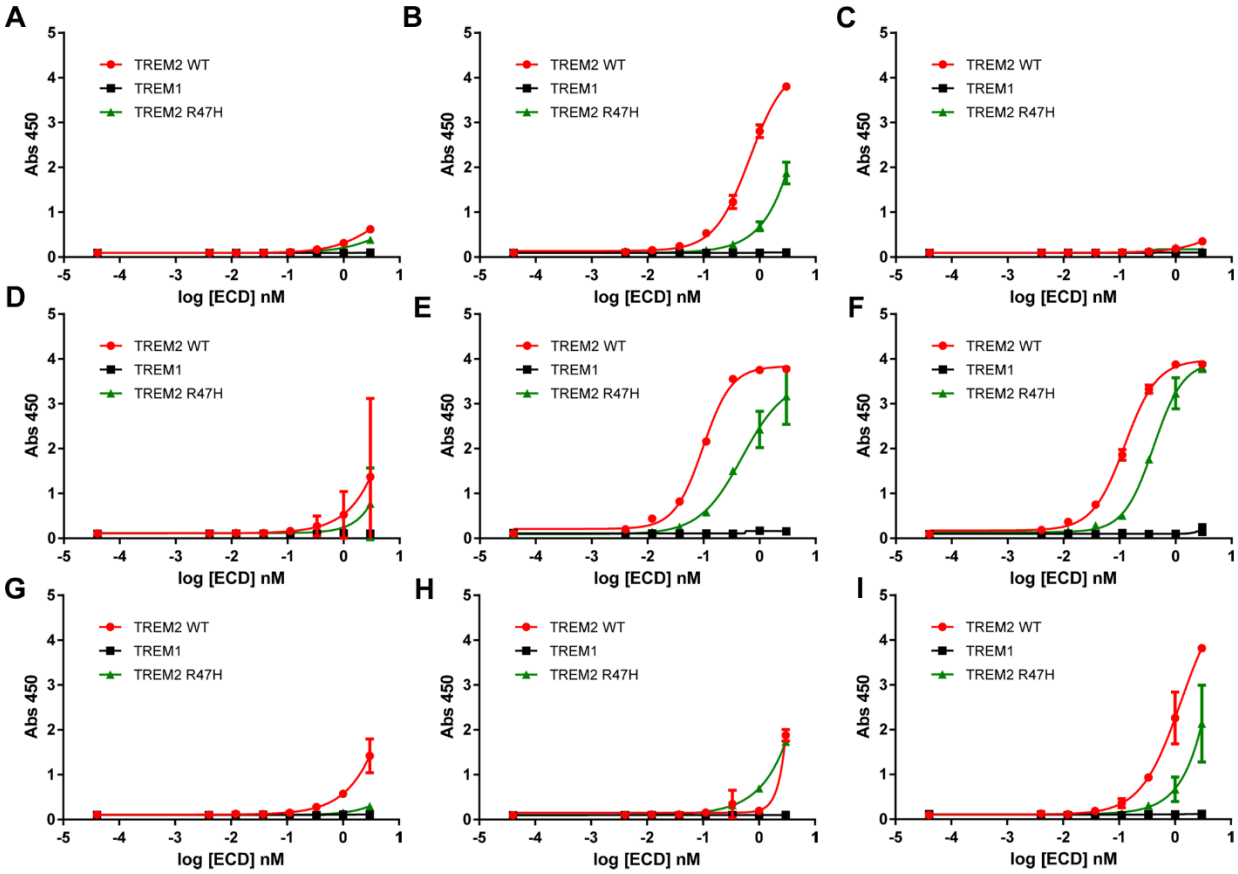

Figure S3. Binding of WT TREM2-ECD, R47H TREM2-ECD and WT TREM1-ECD to various lipids. (a) lysophosphatidic acid (LPA), (b) oxidized phosphatidylserine (OxPS), (c) phosphatidic acid (PA) (d) phosphatidylcholine (PC) (e) phosphatidylethanolamine (PE) (f) phosphatidylglycerol (PG) (g) phosphatidylinositol 4-phosphate (PI4P) (h) sphingolipid (Sph) (i) cardiolipin (CL).

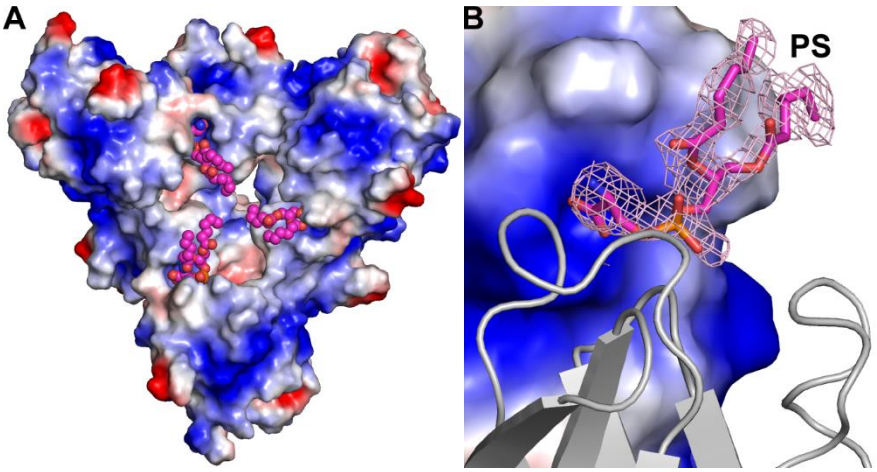

Figure S4. (a) Electrostatic surface diagram of PS-bound WT TREM2 trimeric complex with PS depicted in fuchsia spheres. (b) $F_O - F_C$ electron density omit map for phosphatidylserine with the best electron density bound in TREM2 trimer 'pore', contoured to 3.0σ. PS is depicted in fuchsia sticks. Electron density is observed for the phosphoserine head group but electron density for the hexanoyl tails is variable between molecules, indicating a high degree of mobility.